Reconsiderations of the species status of some South American Planarians (Platyhelminthes: Tricladida: Paludicola)

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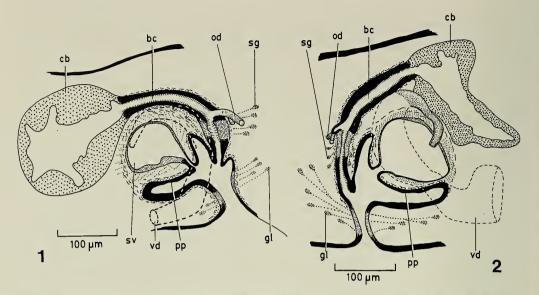
Abstract.—Reexamination of the type material of *Girardia veneranda* (Martins, 1970) revealed that this nominal species is very similar to *G. chilla* (Marcus, 1954). Therefore, *G. veneranda* is determined to be a junior synonym of *G. chilla*. For the first time a morphological description is provided of specimens collected in 1970 near Buenos Aires and identified as *G. anceps* (Kenk, 1930). The features of these *G. anceps* specimens conform in many details with Böhmig's (1902) redescription of this species. The material examined is compared also with more recent descriptions, from other localities, of *G. anceps* and with taxonomically related species.

Adequate assessment of the world's biodiversity depends on up to date knowledge on the diversity of species, their characteristics, and their distribution. The literature often features species descriptions and identifications that are poorly documented or cannot stand scrutiny. In each of these circumstances published results need to be checked on available, original material. This paper analyzes and discusses the species status of several South American planarians from the genus Girardia Ball, 1974. Planarians exemplify the situation that species are frequently characterized by a unique combination of diagnostic features instead of showing apomorphic characters (see Sluys 1991 for a review on species concepts). The variability that is encountered in these softbodied animals has made species recognition in Girardia often very complex and difficult.

De Vries & Sluys (1991) analyzed the phylogenetic relationships between the various genera and subgenera within the Dugesiidae Ball, 1974. They showed that the subgenera *Dugesia* Girard, 1850, *Schmidtea* Ball, 1974, and *Girardia* Ball, 1974, are separate phylogenetic groups and that *Du*- gesia is not closely related to the other two subgenera. Therefore, they proposed to raise each of these subgenera to the rank of genus. The postulated apomorphies for the genus *Girardia* are a pigmented pharynx and a high triangular head shape with pointed auricles, being features that characterize many paludicolans from North America, Central and South America, and the Caribbean.

The present paper for the first time provides morphological evidence supporting the suggestions of Ball (1974) and Kenk (1974) that *G. veneranda* (Martins, 1970) from the State of São Paulo might be identical with *G. chilla* (Marcus, 1954) from southern Chile.

In 1970 Durán-Troise & De Lustig published the chromosome portrait of *Girardia anceps* (Kenk, 1930) from Buenos Aires, Argentina. Unfortunately, these authors did not provide a taxonomic description supporting the identification of their material. Apart from the fact that the identification of Durán-Troise & De Lustig needs proper documentation, examination of their material is also interesting in the light of Moretto's (1991) remark that *G. anceps* does



Figs. 1–2. *Girardia chilla.* 1, MZUSP 5829–5830. Sagittal reconstruction of the copulatory apparatus. bc: bursal canal; cb: copulatory bursa; gl: gland; od: oviduct; pp: penis papilla; sg: shell gland; sv: seminal vesicle; vd: vas deferens; 2, MZUSP 5819–5822. Sagittal reconstruction of the copulatory apparatus. bc: bursal canal; cb: copulatory bursa; gl: gland; od: oviduct; pp: penis papilla; sg: shell gland; vd: vas deferens.

not occur near Buenos Aires. Recently, Puccinelli & Deri (1991) also described the chromosomes of *G. anceps* from Buenos Aires, unfortunately without an anatomical description of their specimens or mentioning the deposition of voucher specimens.

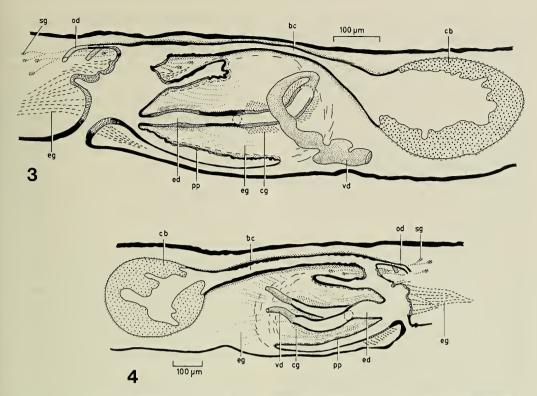
Taxonomic Section

Girardia veneranda (Martins, 1970) Figs. 1, 2

Material examined.—Although the major part of the material studied by Martins (1970) was examined, the present re-description is based mainly on the following preparations [housed in the Museu de Zoologia, Universidade de São Paulo, Seção Helmintologia (MZUSP)]: 5813–5816 (sagittal sections on 4 slides; slide no. 5813 missing), 5817–5818 (sagittal sections on 2 slides), 5819–5822 (sagittal sections on 4 slides), 5823–5825 (transverse sections on 3 slides), 5826–5828 (sagittal sections on 3 slides), 5831–5830 (sagittal sections on 2 slides), 5831–5834 (horizontal and transverse sections on 4 slides). Description.—In the sectioned material no pigmentation appears to be present underneath the pharynx epithelium. The mouth opening is at about one-third of the distance between the hind wall of the pharyngeal pocket and the root of the pharynx.

The testes are principally situated dorsally, but large testes in the posterior end of the body may occupy the entire dorso-ventral space; the follicles extend from directly posterior to the ovaries to almost the posterior body end. The paired ovaries lie at a short distance behind the brain.

The vasa deferentia, which form well developed false seminal vesicles, recurve at about the level of the gonopore and open separately into the antero-lateral portions of the intrapenial seminal vesicle. This spherical or irregularly shaped seminal vesicle occupies most of the penis bulb and papilla. The wall of the vesicle is lined with a nucleate epithelium that is pierced by numerous openings of highly abundant cyanophilous glands, which lie within the penis bulb as well as directly outside of the bulb.



Figs. 3–4. *Girardia anceps.* 3, ZMA V. Pl. 871.5. Sagittal reconstruction of the copulatory apparatus. bc: bursal canal; cb: copulatory bursa; cg: cyanophilic glands; ed: ejaculatory duct; eg: erythrophilic glands; od: oviduct; pp: penis papilla; sg: shell gland; vd: vas deferens; 4, ZMA V. Pl. 871.1. Sagittal reconstruction of the copulatory apparatus. bc: bursal canal; cb: copulatory bursa; cg: cyanophilic glands; ed: ejaculatory duct; eg: erythrophilic glands; ed: ejaculatory duct; eg: erythrophilic glands; od: oviduct; pp: penis papilla; sg: shell gland; vd: vas deferens.

The secretion is also abundantly present in the seminal vesicle.

The penis papilla is a stubby cone, covered with a nucleate epithelium which is underlain with a layer of circular and longitudinal muscle, successively.

The copulatory bursa is situated directly anterior to the penis bulb. From the bursa, the bursal canal curves smoothly towards the postero-dorsal portion of the atrium; the canal is lined with cuboidal cells bearing long cilia. The canal is surrounded by a layer of intermingled circular and longitudinal muscle. The two oviducts fuse to form a very short common oviduct that immediately communicates with the bursal canal. Ventral to the opening of the common oviduct the bursal canal receives the erythrophilous secretion of shell glands. Another set of eosinophilous glands discharges their secretion into the gonopore.

Girardia anceps (Kenk, 1930) = Planaria dubia Borelli, 1895 Figs. 3, 4

Material examined.—The material examined consists of the material collected and described by Durán-Troise & De Lustig (1970) but identified by Prof. Benazzi. This material consists of the following series of slides, now housed in the Zoological Museum, Amsterdam (ZMA): V. Pl. 871.1: sagittal sections of one animal on 3 slides; V. Pl. 871.2: sagittal sections on 2 slides; V. Pl. 871.3: sagittal sections on 2 slides; V. Pl. 871.4: sagittal sections on 2 slides; V. Pl. 871.5: sagittal sections on 4 slides;

V. Pl. 871.6: transverse sections on 4 slides;V. Pl. 871.7: horizontal sections on 3 slides;V. Pl. 871.8: sagittal sections on 1 slide.

Description.—The slides revealed that the animals are pigmented on dorsal and ventral body surface. The sections also unequivocally showed the pharynx to be pigmented. The mouth opening is located at the hind end of the pharyngeal pocket.

The numerous small testes are situated ventrally, from directly behind the ovaries extending to almost the posterior body end. The ovaries are located at some distance behind the brain.

The vasa deferentia recurve at the level of the penis bulb, penetrating it from the lateral sides. Within the bulb the ducts expand in diameter and within the penis papilla they fuse to form a broad ejaculatory duct. The intrapenial parts of the vasa deferentia and the ejaculatory duct are lined with a well developed, nucleate epithelium and are surrounded by a thin layer of circular muscle. The penis papilla is lined with a flat, infranucleate epithelium. Ejaculatory duct and penis epithelium are pierced by ducts of numerous eosinophilic glands, while cyanophilous glands open into the intrabulbar seminal vesicles.

The narrow male atrium communicates with the common atrium; there is no distinct female atrium. The atria are lined with an infranucleate epithelium. Especially the posterior wall of the common atrium is pierced by openings of numerous eosinophilic glands; the glandular elements being broadly distributed in the surrounding parenchyma.

The bursal canal is more or less "angled"; the short section of the canal that sharply turns towards the dorsal part of the atrium receives the openings of erythrophilic shell glands. The bursal canal is lined with an infranucleate, ciliated epithelium. The musculature of the bursal canal either consists predominantly of circular muscles (as in preparations V. Pl. 871.1 and 871.5) or contains intermingled circular and longitudinal fibres (as in preparation V. Pl. 871.3). The infranucleate oviducts join to form a very short common oviduct that opens into the rear wall of the bursal canal, just dorsally to the angled section of the canal.

In specimen V.Pl.871.1 remnants of a spermatophore project from the ejaculatory duct through the gonopore.

Discussion

The above description of *G. veneranda* differs in some important details from that given by Martins (1970). Although she described the vasa deferentia as opening "... into a single and spacious true seminal vesicle ...," the illustration of the copulatory apparatus (Martins 1970, fig. 5) seems to indicate that the ducts open into a sort of intrapenial papilla. However, this suggestion of an intrapenial papilla merely results from an invagination or fold of the irregularly shaped seminal vesicle. In reality the vasa deferentia penetrate separately the lateral wall of the vesicle, as correctly depicted in figs. 6 and 7 of Martins.

According to Martins, the oviducts open separately into the bursal canal. However, detailed study of both sagittal and transverse sections revealed that the oviducts fuse to an extremely short common duct. This situation is already indicated in Martin's fig. 8 but was interpreted differently in her fig. 5 and in the text description of the copulatory apparatus.

The present re-analysis of the type material of *G. veneranda* reveals that the morphology of the reproductive apparatus is strikingly similar to that of *G. chilla*, as described by Marcus (1954) and Hyman (1959). Both species show the spacious intrapenial seminal vesicle, receiving the abundant cyanophilous secretion. According to Hyman (1959) eosinophilous glands open into the vesicle, but this does not agree with my observations and those of Marcus (1954). The only difference to be noted between the *G. veneranda* specimens and *G. chilla* is that the latter was described with numerous glands opening through the entire epithelium of the common atrium. In the *G. veneranda* material the glands were not as abundantly present as described by Marcus or Hyman. According to Marcus these glands are cyanophilic but Hyman described them as eosinophilic, which conforms to my observations.

In view of the above, it is here concluded that *G. veneranda* is essentially similar to *G. chilla* and that therefore *G. veneranda* must be considered a junior synonym of *G. chilla*.

Because Borelli's (1895) description of *Planaria dubia* [=G. anceps] is rather superficial and therefore does not allow detailed comparison with the material described in the present paper, it is here accepted that Böhmig's (1902) more comprehensive description applies to Borelli's species. This axiom is open for criticism, of course, since Borelli's material came from Asunción, Paraguay and Böhmig's specimens were collected near Buenos Aires, Argentina (but it must be noted that Böhmig did examine Borelli's preparations). However, the main purpose of the present discussion is to compare my material with (1) Böhmig's account of G. anceps, (2) more recent descriptions of the species, and (3) descriptions of taxonomically related species.

Böhmig mentioned for G. anceps the following characteristics: (1) recurved vasa deferentia, (2) infranucleate epithelium lining the atrium and the penis papilla, (3) circular muscles around ejaculatory duct and intrabulbar parts of the vasa deferentia, (4) cyanophilous glands discharging into these intrabulbar seminal vesicles, (5) ejaculatory duct receiving the secretion of eosinophilous glands, (6) proximal, anterior section of bursal canal funnel-shaped, (7) posterior two-thirds of bursal canal lined with infranucleate epithelium, (8) oviducts opening separately into bursal canal, (9) large number of eosinophilous glands (different from the shell glands) discharging into the atrium, (10) ventral testes, (11) dorsal surface dark brown with a pale mid-dorsal stripe. Böhmig's diagrammatic reconstruction of the copulatory apparatus of his *G. anceps* specimens has been redrawn by Cazzaniga & Curino (1987, fig. 2).

It is evident that the material of Durán-Troise & De Lustig (1970), as described in the present paper, conforms in many details with Böhmig's account of G. anceps, the only possible difference being the openings of the oviducts into the bursal canal. With respect to the openings of the oviducts, however, it must be noted that Böhmig's reconstruction (cf. Cazzaniga & Curino 1987, fig. 2) suggests oviducts opening very closely together into the bursal canal. Furthermore, it is important to note that in one particular Girardia species some specimens may have oviducts opening separately (but closely together) into the bursal canal, whereas in other animals the oviducts fuse to form a very short common oviduct. The same phenomenon was mentioned by Cazzaniga & Curino for their specimens of G. anceps.

Especially striking similarities between our material and Böhmig's account are the infranucleate epithelia of penis papilla, atrium, and bursal canal. It is therefore disturbing that Böhmig found Borelli's material with nucleate linings of penis papilla and atria (this may cast doubt on the identicality of Böhmig's and Borelli's material; neither does Borelli depict the funnelshaped section of the bursal canal where it communicates with the bursa). Cazzaniga & Curino also found in their specimens a bursal canal lined with a nucleate epithelium, but the atrium and penis papilla in these animals were provided with an infranucleate lining epithelium. These differences in nucleate/infranucleate epithelia, as reported by various authors, may simply reflect different states of development of the copulatory apparatus, only fully mature individuals with all their epithelia infranucleate. In Dugesia gonocephala (Dugès, 1830) De Vries (1984) found that nucleation of the bursal canal depends on the state of development.

Apart from the differences mentioned above, the material of Cazzaniga & Curino is essentially similar to that studied by me. They mention also the infranucleate epithelium of the oviducts, which are surrounded by successively a layer of circular muscle and nuclei. In contrast to my material, Cazzaniga & Curino reported the pharynx to be unpigmented.

In view of the above, it is concluded that the specimens from Buenos Aires analyzed by Durán-Troise & De Lustig correspond with Böhmig's account of *G. anceps* from the same locality and also with the material described by Cazzaniga & Curino from Bahía Blanca. This contrasts with Moretto's (1991) conclusion that *G. anceps* has been recorded incorrectly from Buenos Aires.

The specimens that Kawakatsu & Rovasio (1992) used for their redescription of G. anceps differs in the following features from my material, Böhmig's account, and the description by Cazzaniga & Curino: (1) dorsal surface uniform blackish brown, without light mid-dorsal stripe, (2) pharynx unpigmented, (3) mouth opening somewhat anterior of posterior end of pharyngeal pocket (as suggested by their fig. 3B), (4) divided atrium, (5) male and common atrium lined with nucleate epithelium, (6) bursal canal with nucleate epithelium, (7) bursal canal provided with very thick muscle coat. Each of these characters is known to vary between specimens of a single species and it is debatable whether a combination of these features indicates a species that is different from G. anceps and possibly from any other known species of Girardia.

Several authors have discussed the similarities of *G. anceps* with *G. sanchezi* (Hyman, 1959) and *G. festae* (Borelli, 1898) and the species status of these taxa (Hyman 1959, Cazzaniga & Curino 1987, Moretto 1991, Kawakatsu & Rovasio 1992) [recently, Curino & Cazzaniga (1993) argued that the original spelling *G. festae* was correct and that later emendations to *G. festai* were

invalid]. Sluys (1992) considered G. sanchezi to be a junior synonym of G. festae because both were described with abundant glands opening into the common atrium. unicellular glands opening into the bursal canal, and with penial glands discharging through the epithelium of the penis papilla. Moreover, G. sanchezi and G. festae were both described with a pale, longitudinal mid-dorsal stripe. However, the present study shows that several of these features occur also in G. anceps, suggesting that G. festae might in turn be a junior synonym of G. anceps. Although G. anceps and G. festae are similar in several aspects of their reproductive system, G. festae is different in that it has unicellular glands piercing the entire length of the nucleate bursal canal, and nucleate linings of penis papilla and atria.

Thus, the conclusion of the present study is that G. sanchezi is a junior synonym of of G. festae and that G. festae and G. anceps are valid species. This conclusion is different from that reached by Cazzaniga & Curino (1987) and Kawakatsu & Rovasio (1992) that G. sanchezi is a valid taxon. However, the conclusion of these workers was based on a comparison of G. sanchezi with G. anceps, whereas the present study suggests that G. sanchezi is morphologically closer to G. festae. There is one feature that argues against G. sanchezi being a junior synonym of G. festae, thus illustrating the point made in the introduction that species recognition in Girardia can be very complex: G. festae has recurved vasa deferentia (cf. Sluys 1992, fig. 2) while this is not the case in G. sanchezi (cf. Hyman 1959, fig. 5).

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